Taken together, the restricted expression of CD22, its inherent high endocytotic property(s), and the specificity of receptor/ligand interactions conferred by cognate glycans, support the potential use of the described liposome-based CD22-targeted chemotherapy of B-cell lymphomas. However, some caveats bear mention in contemplating this approach clinically. The authors use a sialoside construct containing a specific biphenylcarboxyl modification that increases affinity of the glycan to CD22. At the same time, however, this modification increases affinity to another siglec, Sn (Siglec-1), expressed on macrophages. Uptake by macrophages increases clearance of the liposomes (thereby attenuating potency). There is also the real possibility that competing uptake by Sn could induce adverse effects secondary to tissue macrophage cytotoxicity. In this regard, the authors show that a different biphenyl modification (ie, biphenylacetyl) that binds poorly to Sn displays reduced clearance in vivo, but these constructs also have reduced affinity for CD22. More critically, glycosidases that cleave sialic acid linkages (ie, sialidases) are present in serum[8] and on the surfaces of cells, including lymphocytes and granulocytes.[9,10] Desialylation of the cognate glycan would result in loss of selective targeting to CD22 and yield coincident liposomal uptake by asialoglycoprotein receptors (eg, in hepatocytes and macrophages), with resultant delivery of the chemotherapeutic agent to bystander cells. Despite these considerations, the results presented are both novel and provocative in suggesting that a cognate glycan-based approach exploiting the lectin function of CD22 could be an option to antibody-mediated chemotherapeutic targeting of B-cell malignancies. The Chen et al report certainly provides persuasive logic to broadening the potential applications of their approach. Notably, the well-known myeloid antigen CD33 is also a Siglec (it is Siglec-3), which raises the possibility that myeloid leukemias could also be subject to a cognate glycan-based liposomal chemotherapeutic approach (eg, using liposomes containing daunorubicin and/or cytosine arabinoside). Thus, for hematologic malignancies, the promise of selective delivery of cytotoxic agents with improved therapeutic efficacy and minimal systemic effects may just well be realized by, literally, hitting with an expanded sweet spot.

Conflict-of-interest disclosure: The author declares no competing financial interests. ■

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● ● ● LYMPHOID NEOPLASIA

Comment on Schafer et al, page 4798

MLL: exploring the methylome

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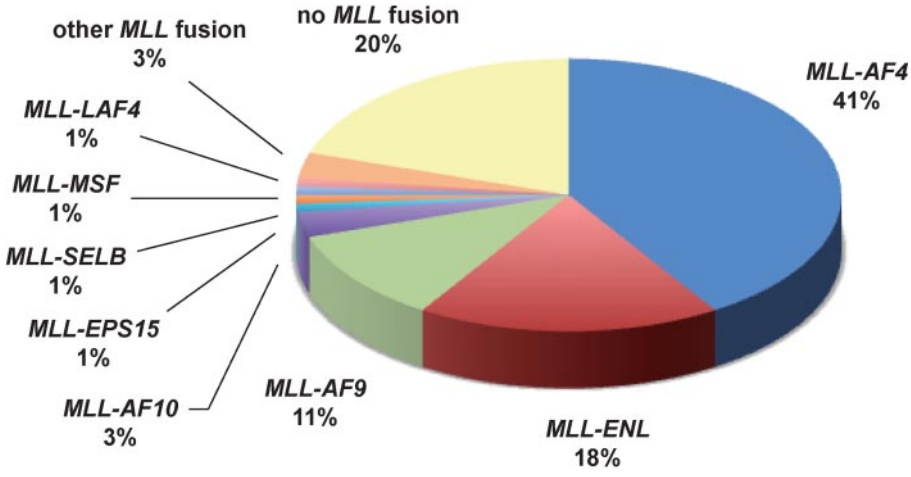
In this issue of *Blood*, Schafer and colleagues report their findings on *MLL*-r infant ALL.[1] They examined global promoter methylation in infants and found hypermethylation in cases with *MLL*-r ALL compared with both the normal and non-*MLL*-r ALL cases. When treating *MLL*-r cell lines with decitabine, they observed a reexpression of silenced genes and a cytotoxic effect. This may open up the prospect of new treatment options for a disease entity with a dismal prognosis.

Hardly any other gene in cancer medicine is involved in as many different chromosomal translocations as the *mixed lineage leukemia* (*MLL*) gene. It has thus far been possible to identify more than 60 different genes dispersed throughout the genome and to cytogenetically characterize another 35 loci that are involved in *MLL* translocations.[2] Around 10% of acute leukemia (AML and ALL) patients carry *MLL* aberrations. These patients have been shown to exhibit a distinct gene expression signature.[3] In acute lymphoblastic leukemia (ALL), *MLL* aberrations are usually associated with an adverse prognosis in all age



Infant ALL has a unique genetic background with approximately 80% of patients carrying *MLL* aberrations (data according to Jansen et al[4]).

groups. More intensified treatment schedules, especially allogeneic transplantation, have brought some improvements in adults. However, these toxic regimens are often considered unacceptable for affected children. While the overall prognosis of pediatric ALL has steadily improved and is considered particularly good in children around the age of 4, the prognosis of infant ALL (ie, younger than 1 year) is still unsatisfactory with event-free survival rates below 50%.[5] The main reason for this unfavorable outcome is the extremely high prevalence (around 80%) of *MLL* translocations in this age group (see figure).

The *MLL* protein has a highly complex function in normal and malignant hematopoiesis. It is part of a large macromolecular nuclear complex that has histone methyltransferase and histone acetyltransferase function and thus epigenetically regulates the expression of several genes and remodels nucleosomes.[6] In particular, several homeobox (*HOX*) genes are regulated via *MLL*. Chromosomal translocations involving *MLL* lead to deregulated *HOX* gene expression and a different transcription pattern.

Schafer et al from Patrick Brown's research group focused on a specific aspect of infant *MLL-r* ALL. They investigated the "epigenome" in these patients, particularly the methylation of promoter islands. They used a specific assay (HELP = *HpaII* tiny fragment enrichment by ligation-mediated PCR).[7] Basically, this assay utilizes the fact that the 2 restriction enzymes *Msp*I and *HpaII* both recognize the same restriction site (5′–C|CGG–3′), but the first enzyme is methylation-insensitive, while the second one is not and will only cut when both cytosines are unmethylated. Thus, these enzymes cut genomic DNA into different fragments depending on its methylation status. The resulting DNA fragments were ligated in a second step to short "linker sequences," amplified by PCR and analyzed on a custom-designed oligonucleotide microarray covering 25 626 *HpaII*-amplifiable fragments located at around 14 000 gene promoters. This analysis revealed significant overall methylation differences between *MLL*-rearranged (*MLL*-r), *MLL*-wild-type and normal samples. *MLL*-r ALL samples were characterized by global CpG island hypermethylation, while healthy controls and *MLL*-wild-type samples showed similar methylation patterns. To validate these findings, Schafer et al performed qRT-PCR on selected genes with a difference in promoter methylation in the previous analysis. Five of 7 genes with hypermethylated promoter regions had a significantly or at least markedly lower expression level. These qRT-PCR data were in accordance with previously published gene expression microarray data. In a further step, ALL cell lines with and without *MLL* fusion genes were investigated and treated with the demethylating agent decitabine. Genes previously found to be promoter-hypermethylated and silenced or underexpressed in *MLL*-r ALL were studied in these cell lines before and after decitabine treatment. Reversal of promoter methylation in *MLL*-r cell lines was revealed by sodium bisulfite treatment and methylation-specific PCR. Quick reexpression of these genes was observed, and this was associated with a cytotoxic effect on the *MLL*-r but not the *MLL*-wt cell lines.

In conclusion, the results reported by Schafer et al reveal interesting aspects of the biology of *MLL*-r infant ALL.[1] However, they should be confirmed by studies involving larger numbers of patients. Several questions remain to be answered, especially concerning the molecular pathways leading to this global hypermethylation. The degree of methylation may also depend on the *MLL* fusion partner, as recently suggested.[8] Nevertheless, there is increasing evidence that hypermethylation plays an important role in infants and perhaps also in older patients with *MLL*-r ALL. This should encourage the initiation of clinical studies investigating the potential benefit of demethylating agents in acute leukemias with *MLL* aberrations.

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● ● ● TRANSPLANTATION

Comment on Coghill et al, page 4914

Leading T cells astray

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In this issue of *Blood*, Coghill and colleagues reveal a critical role of lymphoid CCR7 for the initiation of acute GVHD.[1] The authors report that CCR7 deficiency impairs the initiation of acute graft-versus-host but does not interfere with the desired antitumor effect. Can the interference with this important homing receptor bring the field of allo-HCT one step closer to achieving the ultimate goal of providing a targeted cellular anticancer therapy without the risk of GVHD?

A functioning immune system is highly dependent on interactions between specialized immune cells at the right time in the right place. For this reason, dynamic and coordinated immune cell migration throughout the body and within tissues is a prerequisite for efficient immunosurveillance and tolerance. Immune cell migration is orchestrated by the precise interplay of adhesion molecules and chemokines and their appropriate receptors, and has become the focus of attention in recent years due to the potential to modulate immune cell function for therapeutic purposes.